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Coexistence of Deficiencies of Uroporphyrinogen III Synthase and Decarboxylase in a Patient with Congenital Erythropoietic Porphyria and in His Family

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Summary: A hitherto undescribed dual deficiency of uroporphyrinogen III synthase and uroporphyrinogen decarboxylase was observed in the erythrocytes in a 14 year-old patient who had presented with congenital erythropoietic porphyria since early childhood. Whereas congenital erythropoietic porphyria was metabolically and clinically overt, a hereditary deficiency of uroporphyrinogen decarboxylase was confirmed by family study. The uroporphyrinogen III synthase activity of the propositus was decreased to 26% of the control while his asymptomatic family members had activities between 53–65% of the control. Additionally, the uroporphyrinogen decarboxylase activity was 55–66% of the control in the patient and his family. Family investigations have shown that the two disorders do not consistently segregate together. Although urinary porphyrin excretions of relatives were in the physiological range, the proportion of coproporphyrin isomer I showed a relative increase, which can serve as a biochemical indicator for heterozygous uroporphyrinogen III synthase gene carriers.

Introduction

Porphyrias are mainly hereditary diseases, each one reflecting a partial genetic defect of one of the enzymes in the haem biosynthetic pathway (1). Simultaneous presence of the enzyme deficiencies of two types of hereditary porphyria in one individual and/or in the same family is characteristic of dual porphyrias. These conditions are rare, and have so far been reported only as combinations of acute intermittent porphyria and porphyria variegata, known as the Chester Type (McKusick 176010) (2), acute intermittent porphyria (McKusick 176000) and porphyria cutanea tarda (McKusick 176100) (3, 4), porphyria variegata (McKusick 176200) and porphyria cutanea tarda (5–8), and hereditary coproporphyria (McKusick 121300) with congenital erythropoietic porphyria (McKusick 163700) (9) (fig. 1).

This present report describes a patient with congenital erythropoietic porphyria who was also found to have coincidentally inherited half-normal activity of uroporphyrinogen decarboxylase[1]).

[1]) Enzymes
Porphobilinogen synthase (EC 4.2.1.24)
Porphobilinogen deaminase (EC 4.3.1.8)
Uroporphyrinogen III synthase (EC 4.2.1.75)
Uroporphyrinogen decarboxylase (EC 4.1.1.37)

Patient and Methods

The male patient, born in 1981, developed excessive haemolytic anaemia as a newborn. Photosensitivity became obvious after phototherapy which was indicated because of severe jaundice. Blistering occurred on foot, stomach, chest and forehead, and the healed areas were hypopigmented. Additionally, red coloured urine was observed. Family members were clinically unaffected. During the subsequent years the clinical symptoms of congenital erythropoietic porphyria seen in the patient exacerbated and were accompanied by development of severe osteoporosis, renal and liver siderosis and nephrotic syndrome (10). Further porphyrin studies were performed in 1995.

Porphyrins were determined by high performance thin-layer chromatography in combination with absorption spectrophotometry (11). Urinary and faecal coproporphyrin isomers I/III were analysed as free acids by C_8 reversed phase high performance liquid chromatography in connection with fluorometric detection (12). Porphobilinogen synthase, porphobilinogen deaminase and uroporphyrinogen decarboxylase activities were determined as previously described (3, 14, 15). Uroporphyrinogen III synthase activity in red cell lysates was determined by a coupled enzyme assay adapted from *Tsai* et al. (16) with slight modifications. Incubation was carried out at 37 °C for 15 min in the dark. The reaction was stopped by the addition of 500 µl trichloroacetic acid containing iodine and riboflavin. The latter served as an internal standard. The oxidized uroporphyrin isomers were separated by C_{18} reversed-phase HPLC (RP 18 LiChroCART® 4 · 70 mm, Merck, Darmstadt, Germany) and quantitated by fluorometric detection.

Results

The first pathobiochemical investigation of the patient at the age of 24 days showed elevated porphyrin levels in urine, faeces and blood. Metabolic data are compiled

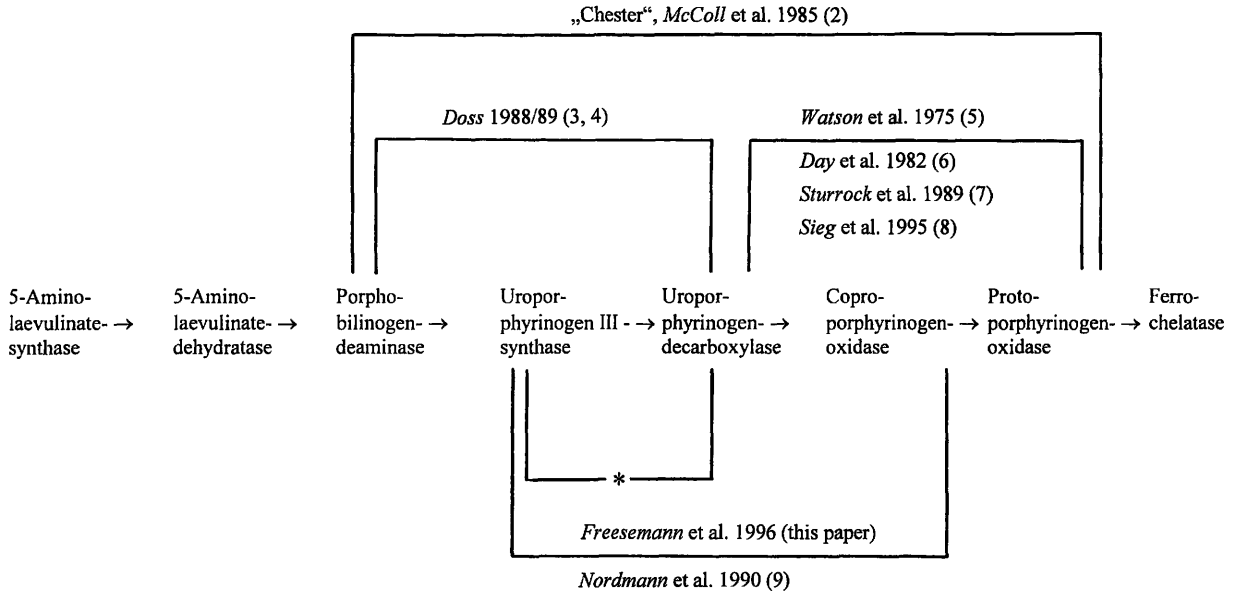

Fig. 1 All the known dual porphyrias and the corresponding defective enzymes in haem biosynthesis (* indicates the dual enzyme deficiency reported in this paper).

in table 1 and 2. Urinary porphyrins consisted mostly of uro- and coproporphyrins, while the predominant faecal porphyrin was coproporphyrin; in each case there was a high predominance of isomer I. These characteristic constellations confirmed the clinical suspicion of congenital erythropoietic prophyria. We found a decreased activity of uroprophyrinogen decarboxylase in erythrocytes, which is not typical of congenital erythropoietic porphyria. We therefore performed metabolic and enzyme studies on both the patient and his parents. Porphyrinaemia and porphyrinuria were exacerbated with clear predominance of uroprophyrin in urine and blood. Enzyme studies again showed a reduced activity of uroporphyrinogen decarboxylase (60% of control) and revealed a dual deficiency with decreased activity of both uroporphyrinogen III synthase (26% of control), as seen in congenital erythropoietic porphyria, and uroporphyrinogen decarboxylase, as seen in porphyria cutanea tarda (tab. 3).

His parents had decreased uroporphyrinogen III synthase activities at an intermediate level (around 60% of control) and, notably, both parents also showed decreased activity of uroporphyrinogen decarboxylase (tab. 3), i.e. both parents were double heterozygotes (fig. 2). Red cell porphobilinogen synthase and deaminase activities were normal in the patient and his family (data not shown). No significant increases in porphyrins were found in urine, faeces or blood of family members. However, the urinary coproporphyrins of family members showed an increase in the isomeric series I (tab. 1). His sister was also found to be doubly heterozygous for the trait of congenital erythropoietic porphyria and porphyria cutanea tarda, as shown by enzyme activities and a slight increase in the proportion of urinary coproporphyrin I (tabs. 1 and 3).

Discussion

A male child with a hitherto undescribed double enzyme deficiency of uroporphyrinogen III synthase and uroporphyrinogen III decarboxylase, associated with the clinical feature of congenital erythropoietic porphyria, is presented. His parents were found to be heterozygous for both the trait of congenital erythropoietic porphyria and the trait of porphyria cutanea tarda. Therefore, the

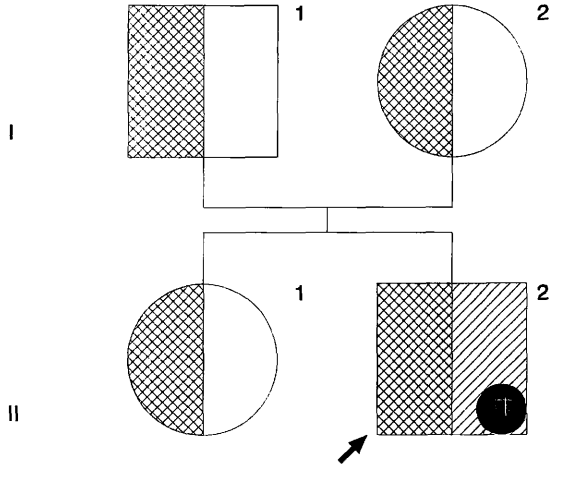

Fig. 2 Pedigree of family with dual enzyme deficiency. ◐ ◧ heterozygous deficient of uroporphyrinogen III synthase and uroporphyrinogen decarboxylase activities, ▩ heterozygous deficient of uroporphyrinogen decarboxylase activity and homozygous deficient of uroporphyrinogen III synthase activity, ● clinical manifestation of congenital erythropoietic porphyria; ↗ propositus.

Tab. 1 Urinary and faecal excretion of porphyrins by the patient and his family

	Urinary porphyrins (nmol/24 h)						Faecal porphyrins (nmol/g)			
	Uro-	Hepta-carboxy-	Hexa carboxy-	Penta-carboxy	Copro-	Ratio copro- I/III	Total	Copro-	Ratio copro- I/III	Proto-	Total
Patient in 1981	413	10	2	33	425	90/10	883	96	97/3	18	122
Patient in 1995	122,449	6,312	2,727	6,162	43,440	96/4	181,090	19,355	96/4	25	19,380
Father	14	2	1	3		93/7	100	11	78/22	53	64
Mother	12	2	1	4	122	63/37	136	6	72/28	50	56
Sister	17	3	2	4	133	48/52	152	8	64/36	48	56
Reference range	4–30	0–4	0–3	0–6	21–120	17–31/69–83	25–165	5–37	60–75/25–40	21–151	30–224

uroporphyrinogen decarboxylase deficiency of the patient was inherited. Low erythrocyte uroporphyrinogen decarboxylase activity is observed in porphyria cutanea tarda type II, the familiar type (1, 17). The genes encoding uroporphyrinogen III synthase and uroporphyrinogen III decarboxylase are located on different chromosomes. The human congenital erythropoietic porphyria gene has been mapped to chromosome 10 q25 · 2 − q26 · 3 (18−21). The gene for uroporphyrinogen decarboxylase has been assigned to the region 1 pter-p34 of the human chromosome 1 (17). This suggests that this case of dual enzyme deficiency results from the incidental genetic transmission of two independently occurring gene defects, as confirmed by the family investigation (tab. 3). The patient received a uroporphyrinogen III synthase gene defect from each parent and a uroporphyrinogen decarboxylase gene defect from only one parent (fig. 2). From this it can be concluded that these two defects do not segregate together. Hepatoerythropoietic porphyria, the homozygote form of porphyria cutanea tarda, can be excluded by the enzymatic and metabolic data given before.

The propositus developed the clinical syndrome of congenital erythropoietic porphyria shortly after birth. Coinheritance of uroporphyrinogen decarboxylase deficiency does not appear to have modified the phenotype of congenital erythropoietic porphyria. Whereas congenital erythropoietic porphyria follows an autosomal recessive trait of inheritance, genetically determined porphyria cutanea tarda follows an autosomal dominant trait of inheritance, though with a low clinical penetrance. Lack of modification of the phenotype by uroporphyrinogen decarboxylase deficiency is not unexpected, since in most of those known to have inherited this deficiency it is clinically and metabolically latent (17). Furthermore congenital erythropoietic porphyria and porphyria cutanea tarda are both porphyrin accumulation disorders in which skin photosensitivity and chronic photodermatosis are main clinical symptoms (22, 23). Cutaneous symptoms of congenital erythropoietic porphyria are much more severe than those of porphyria cutanea tarda; in addition, porphyria cutanea tarda usually becomes clinically overt in adult life (1, 24). Therefore, the clinical consequences of the uroporphyrinogen decarboxylase gene defect would be covered by the clinical features of congenital erythropoietic porphyria.

The porphyrin metabolite pattern of the patient reflected uroporphyrinogen III synthase deficiency. A slight metabolic expression of uroporphyrinogen decarboxylase deficiency became obvious from the higher proportion (1 : 0.3) of urinary uroporphyrin, in comparison with other congenital erythropoietic porphyria cases (1 : 0.6; n = 7). Additionally, reinvestigation after 14 years revealed an increase in the proportion of uroporphyrin from 47% to 68% of total urinary porphyrins.

Tab. 2 Blood porphyrin profiles of the family

	Plasma porphyrins (nmol/l)					Erythrocyte porphyrins (nmol/l)		
	Uro-	Hepta-carboxy-	Copro-	Proto-	Total	Uro-	Proto-	Total
Patient in 1981	432	13	581	18	1058	61	2065	2126
Patient in 1995	4,032	229	1,438	140	6,191	11,724	5,162	16,886
Father	2	1	0.5	14	18	1	267	269
Mother	1	1	1	12	15	1	249	250
Sister	1	0.5	1	11	14	1	338	340
Reference range	0–1	0–1	0–3	0–15	0–19	0–1	90–640	90–670

Tab. 3 Enzyme activities in the erythrocytes of the patient and his relatives (cf. pedigree in fig. 2) in % of control

Enzyme	I_1 Father	I_2 Mother	II_1 Sister	II_2 Patient	Reference range (nkat/l) [$\bar{x} \pm$ SD (n)]
Porphobilinogen deaminase	74	97	93	83	22 ± 3 (68)
Uroporphyrinogen III synthase	65	59	53	26	702 ± 62 (10)
Uroporphyrinogen decarboxylase	57	55	66	60	2.8 ± 0.4 (35)

The uroporphyrinogen III synthase deficiency is clinically overt in the homozygous state, but clinically silent in the heterozygous condition. Total porphyrin excretion as well as urinary uro- and coproporphyrin excretion of the gene carriers (parents and sister) were normal. However, the relative increase of coproporphyrin isomer I indicates intermediate uroporphyrinogen III synthase deficiency. This isomer III/I inversion is a metabolically intriguing sign and can serve as a biochemical indicator of heterozygotes. Uroporphyrinogen decarboxylase deficiency is clinically silent in family members. Urinary uro- and heptacarboxyporphyrin excretion are normal, indicating that, at the time of investigation, the disposition for porphyria cutanea tarda was in the genetic phase without metabolic consequences (22). All types of porphyria cutanea tarda can be triggered by liver diseases, alcohol and oestrogens (1). Obviously, the parents were not exposed to such triggering factors. The observation that a porphyria cutanea tarda gene carrier can show a decreased enzyme activity without metabolic and clinical consequences is well known (17, 25).

This combination of congenital erythropoietic porphyria and hereditary coproporphyria (9), is also the second report of a dual enzyme deficiency in which one disorder is autosomal recessive and the other is autosomal dominant.

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